Poster II-31

High-Throughput Image Cytometry of Subcellular Signals

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Assay analysis at the subcellular level for large cell populations is enabled by high-throughput microscopy (HTM), which provides the automation, image resolution and image processing required for target localization and pathway-component visualization within cellular signaling networks. The impact of technological advances offered by HTM in combination with powerful data mining tools enabled by per cell analysis of entire populations and sub-populations has resulted in automated assays carried out at subcellular resolution in microtiter plate screening formats including subcellular compartment or organelle translocation, aggregate formation, and morphology changes.

Cell-based imaging HTM enables high-resolution analysis of multiple target-protein-dependent parameters. The challenge of providing algorithms that convert raw image data into parameter read-outs for quantification of various biological phenomena is presented in the context of subcellular cytometric assays, like NFκB translocation, GPCR vesicle or pit formation (shown in Figure 1), and differential foci formation in intranuclear AR-trafficking. These and other applications require analysis and retrieval of each of the 1,000s to 100,000s of imaged cell objects in each plate. Data mining research challenges include an integrated database for linking data to corresponding cell images through a simple software interface that enables efficient plotting, searching, and review of data-linked images. Drug discovery is just one example of how high throughput automation of subcellular information over large cell populations opens a new world of cellular discovery. As high throughput automation of high-resolution microscopy images evolves, previously overwhelming numbers of cellular and subcellular details will be increasingly integrated into more organized and useful information for cell-based sciences.

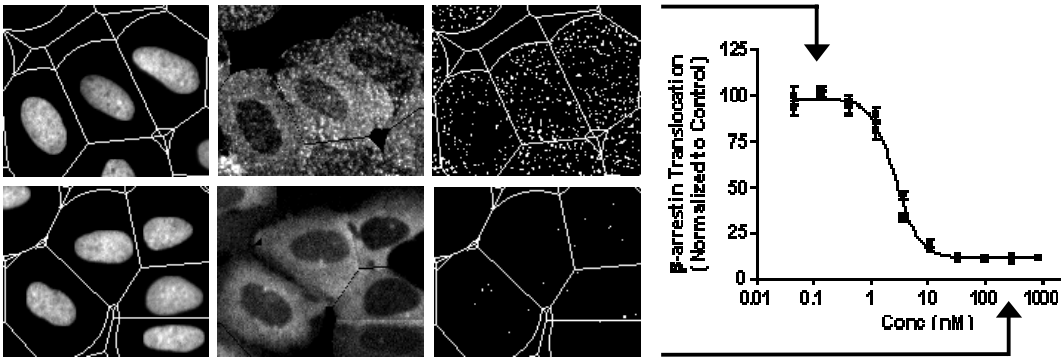

Figure 1: GPCR assay analysis including partitioning the cellular regions (left) and the inhibitory dose response curve (right). The images show GPCR assay analysis with pit formation examples in the top row and inhibition in the bottom row. The left column shows the nuclei stained with Hoechst, which are used to assign the object areas. The middle column shows the corresponding GFP channel images, and the right column demonstrates the binary masks for the formed pits. EIDAQ™ 100 instrument and CytoShop™ software from Q3DM Inc. and plate preparation courtesy of Norak Biosciences Inc.

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